

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims:

WHAT IS CLAIMED:

1. (CANCEL): A nanoknife comprising:

- a solid blade body having a roughly quadrangular base surface, two opposite roughly triangular side surfaces, and two opposite roughly regular trapezoid side surfaces, said side surfaces meeting to form an edge; and
- a moveable mounting to which said base is affixed thereby allowing said blade to be selectively positioned to make a desired manipulation.

2. (CANCEL): The knife of claim 1, wherein said edge is approximately 100 microns long.

3. (CANCEL): The knife of claim 1, wherein said edge is less than approximately 10 microns long.

4. (CANCEL): The knife of claim 1, wherein said edge is less than 200 microns long.

5. (CANCEL): The knife of claim 1, wherein said edge is approximately 100 microns from said base surface.

6. (CANCEL): The knife of claim 1, wherein said blade body is manufactured according to a method for manufacturing an atomic force microscope point but providing extension of said point into said edge.

7. (CANCEL): The knife of claim 1, wherein said moveable mounting comprises a flexible cantilever.

8. (CANCEL): The knife of claim 1, wherein said solid body is formed from a transparent material and mounted such that an object to be manipulated can be viewed during positioning and manipulation.

9. (CANCEL): The knife of claim 1 further comprising:

a MEMS axon knife module comprising:

a plurality of electrical contacts;

glass insulation;

one or more heated beams;

two thermal actuators; and

wherein said knife module comprises a flexible knife frame.

10. (ORIGINAL): A method of repairing a damaged nerve in a living organism comprising:

selecting one or more axons in said damaged nerve;

harvesting a donor axon segment;

positioning said donor axon segment at a severed location of a selected axon; and

inducing fusion of said donor axon segment.

11. (ORIGINAL): The method of claim **10** further comprising:

cutting one or more ends of said donor axon and/or said selected axon.

12. (ORIGINAL): The method of claim **10** further comprising:

wherein said positioning comprises:

applying a dielectrophoresis signal in proximity of one or more of said donor axon or said selected axon.

13. (ORIGINAL): The method of claim **10** further comprising:

wherein said inducing comprises:

applying a electric signal in proximity of one or more of said donor axon or said selected axon.

14. (ORIGINAL): The method of claim **10** further comprising:

digesting one or more nerve portions to allow manipulation of individual axons.

15. (ORIGINAL): The method of claim **10** further comprising:

using a MEMS axon surgical platform enabling precise manipulation of axons of less than one and up to a few microns in diameter.

16. (ORIGINAL): The method of claim **10** wherein said living animal comprises a human.

17. (ORIGINAL): The method of claim **10** wherein said living animal comprises a mammal.

18. (ORIGINAL): The method of claim **15** further wherein:

said MEMS axon surgical platform enables manipulation of axons directed by a human surgeon.

19. (ORIGINAL): A method of repairing a damaged nerve in a living organism comprising:

selecting one or more severed axons in said damaged nerve;

positioning one or more of said selected severed axons in close proximity to one or more

corresponding severed axons on another side of said damaged nerve; and

inducing fusion of axons placed in proximity;

said one or more corresponding severed axons are not necessarily matched to axon

segments to which they were attached before becoming severed; and

cutting said one or more severed axons prior to said inducing..

20. (CANCEL): The method of claim **19** further wherein:

said one or more corresponding severed axons are not necessarily matched to axon segments to which they were attached before becoming severed.

21. (CANCEL): The method of claim **19** further comprising:

cutting said one or more severed axons prior to said inducing.

22. (CANCEL): A method of constructing a three-dimensional microstructure comprising:

constructing a plurality of separable planar components from a planar substrate using one

or more microfabrication techniques, wherein up to all of said plurality includes one or

more interlock structures; and

assembling said separable planar components into a three-dimensional microstructure.

23. (CANCEL): The method of claim **22** further wherein:

said planar components comprise:

two substantially identical top and bottom components; and
four substantially identical side components.

24. (CANCEL): The method of claim **22** further wherein:

said three-dimensional microstructure is smaller than approximately one cubic millimeter.

25. (CANCEL): The method of claim **22** further wherein:

said three-dimensional microstructure is smaller than approximately five cubic millimeters.

26. (CANCEL): The method of claim **22** further wherein:

said three-dimensional microstructure is smaller than approximately 100 cubic millimeters.

27. (CANCEL): The method of claim **22** further wherein:

said planar components comprise:

a plurality of space-frame components that can be assembled into a three-dimensional structure for holding functional components; and

one or more modular functional components able to be arranged in said three-dimensional structure.

28. (CANCEL): The method of claim **27** further wherein:

said one or more modular functional components comprise:

one or more nanoknives;

one or more actuators for moving said nanoknives; and

one or more effector electrodes for moving cells and/or portions of cells;

and said three-dimensional microstructure comprises a microsurgery platform.

29. (CANCEL): The method of claim **28** further comprising:

attaching said three-dimensional microstructure to a carrier platform for positioning by a manipulator.

30. (CURRENTLY AMENDED): ~~The method of claim 28 further comprising:~~ A method of constructing a three-dimensional microstructure comprising:

constructing a plurality of separable planar components from a planar substrate using one or more microfabrication techniques, wherein up to all of said plurality includes one or more interlock structures; and

assembling said separable planar components into a three-dimensional microstructure; said planar components comprising:

a plurality of space-frame components that can be assembled into a three-dimensional structure for holding functional components; and

one or more modular functional components able to be arranged in said three-dimensional structure;

said one or more modular functional components comprising:

one or more nanoknives;

one or more actuators for moving said nanoknives; and

one or more effector electrodes for moving cells and/or portions of cells;

said three-dimensional microstructure comprises a microsurgery platform;

arranging said three-dimensional microstructure in a surgical frame, said surgical frame comprising one or more gaskets for holding one or more nerves to be repaired;

arranging in said three-dimensional microstructure and/or said carrier platform and/or said surgical frame one or more microfluidic channels for delivering reagents; and

arranging in said three-dimensional microstructure and/or said carrier platform and/or said surgical frame one or more waveguides to enable optical monitoring, visualization, and /or use of light sources.

31. (CANCEL): The method of claim 30 further comprising:

arranging in said three-dimensional microstructure and/or said carrier platform and/or said surgical frame one or more microfluidic channels for delivering reagents.

32. (CANCEL): The method of claim 30 further comprising:

arranging in said three-dimensional microstructure and/or said carrier platform and/or said surgical frame one or more waveguides to enable optical monitoring, visualization, and /or use of light sources.

33. (CANCEL): A three-dimensional microsurgery platform comprising:

a nanoknife;
one or more actuators for moving said nanoknife up and down; and
one or more electrodes positioned near said nanoknife for delivering controlled electrical signals in proximity to said nanoknife.

34. (CURRENTLY AMENDED): ~~The device of claim 33 further wherein:~~ A three-dimensional microsurgery platform comprising:

a nanoknife;
one or more actuators for moving said nanoknife up and down;
one or more electrodes positioned near said nanoknife for delivering controlled electrical signals in proximity to said nanoknife, further wherein
said one or more electrodes are connectable to an external signal source thereby able to deliver electric signals to orient and/or move cells and/or cell components;
said one or more electrodes are connectable to an external signal source thereby able to deliver electric signals to induce fusion of cells and/or cell components.

35. (CANCEL): The device of claim ~~33~~34 further wherein:

said one or more electrodes are connectable to an external signal source thereby able to deliver electric signals to induce fusion of cells and/or cell components.

36. (CURRENTLY AMENDED): The device of claim ~~33~~34 further wherein:

one or more of said electrodes comprise an electrode array, each having at least two addressable pin/probe and/or plate electrodes.

37. (CURRENTLY AMENDED): The device of claim ~~33~~34 further wherein:

said one or more electrodes have at least three separable modes of operation comprising:
a positioning mode for moving cells and/or cell components;

a joining mode for moving two or more cells and/or cell components in proximity to each other; and

a fusion mode for inducing fusion of said cells and/or cell components.

38. (CURRENTLY AMENDED): The device of claim ~~33~~34 further wherein:

said microsurgery platform is smaller than approximately one cubic millimeter

39. (CANCEL): The device of claim **33** further wherein:

said microsurgery platform is smaller than approximately one hundred cubic millimeters.

40. (CANCEL): The device of claim **33** further comprising:

a plurality of pins and/or rods for affixing said microsurgery platform to a manipulator.

41. (ORIGINAL): A system able to repair nerves in living animals comprising:

a surgical frame container able to isolate a damaged nerve portion in a living subject;

said surgical frame comprising:

one or more nerve gaskets for holding nerve portions in said frame container;

one or more wave guides for providing illumination within said frame container;

a plurality of fluid inflow and outflow channels able to maintain a desired medium with desired properties in said surgical frame container;

an opening able to receive a three-dimensional microsurgery platform and allowing access to said platform to a manipulator.

42. (ORIGINAL): A method of manipulating small objects using electrical energy in a micro-manipulation system comprising:

precisely positioning arrangements of micro-electrodes near to said objects; and

applying electrical signals precisely to particular electrodes to effect precise movements and/or manipulations.

43. (ORIGINAL): The method of claim **42** wherein said small objects comprise one or more cells and/or cell components.

44. (CURRENTLY AMENDED): The method of claim **42** wherein said precisely positioning of arrangements of electrodes comprises positioning an addressable grid of electrodes above and/or below said objects and further comprising:

observing said small objects in relation to said addressable grid of electrodes;

selecting one or more electrodes that will effect a desired movement of said small object;

and

applying a predetermined energy signal to said one or more electrodes; and further comprising:

observing an induced movement of said small objects in relation to said addressable grid of electrodes;

selecting an additional one or more electrodes that will effect a desired further movement of said small object; and

applying a predetermined energy signal to said additional one or more electrodes.

45. (CANCEL): The method of claim **44** further comprising:

observing said small objects in relation to said addressable grid of electrodes;

selecting one or more electrodes that will effect a desired movement of said small object;

and

applying a predetermined energy signal to said one or more electrodes.

46. (CANCEL): The method of claim **44** further comprising:

observing an induced movement of said small objects in relation to said addressable grid of electrodes;

selecting an additional one or more electrodes that will effect a desired further movement of said small object; and

applying a predetermined energy signal to said additional one or more electrodes.

47. (ORIGINAL): The method of claim **42** wherein said small objects comprise axons.

48. (ORIGINAL): A method of manipulating an axon segment comprising:

using dielectrophoresis (DEP) to move said axon segment in a non-homogeneous electrical field.

49. (ORIGINAL): The method of claim **48** further comprising:

using dielectrophoresis (DEP) to align axon segments.

50. (ORIGINAL): The method of claim **48** further wherein said electric field has frequencies above about 5 kHz.

51. (CURRENTLY AMENDED): The method of claim **48** further comprising:

placing said axons in a medium with a conductivity selected to influence the magnitude and direction of DEP force; and

placing said axons in a medium modified to optimize physiological conditions of axons during DEP, and containing additives to promote one or more of axon fusion, visualization of axons, or other manipulations on axons.

52. (CANCEL): The method of claim **48** further comprising:

placing said axons in a medium modified to optimize physiological conditions of axons during DEP, and containing additives to promote one or more of axon fusion, visualization of axons, or other manipulations on axons.

53. (ORIGINAL): The method of claim **48** further comprising:

adjusting a frequency of an applied voltage to precisely control the magnitude and direction of DEP.

54. (ORIGINAL): A method of determining frequency parameters for manipulating an axon segment comprising:

selecting a desired electrode design;

selecting a desired fluidic medium with a conductivity different from an interstitial axon conductivity;

determining a first DEP crossover frequency along a short axis of an axon of a type to be moved;

determining a second DEP crossover frequency along a long axis of an axon of a type to be moved; and

selecting a frequency between said first and said second DEP crossover frequencies such that attractive DEP forces occur along a length of said axon segment and repulsive DEP forces occur in a direction perpendicular to said length;
wherein said first and second crossover frequencies are frequency points at which a force on said axon segment transitions from an attractive force to a repulsive force.

55. (ORIGINAL): The method of claim **54** further comprising:

for a particular axon segment, using a pin/probe and plate electrode pair such that a region of strongest field is towards the pin electrode thereby stretching an axon towards said pin/probe electrode.

56. (ORIGINAL): The method of claim **54** further comprising:

for a particular axon segment, using an addressable grid of electrodes to addressably select electrode pairs to move an axon segment as desired.

57. (ORIGINAL): The method of claim **55** further comprising:

for at least two axon segments, using a pin/probe electrode and multiple plate electrodes such that multiple axons are stretched towards said pin/probe electrode.

58. (ORIGINAL): The method of claim **55** further comprising:

using multiple pin/probe and plate electrodes to move and/or align different segments of axons.

59. (ORIGINAL): The method of claim **55** further wherein:

said pin/probe is an energized electrode.

60. (ORIGINAL): The method of claim **55** further wherein:

said pin/probe is a non-energized dielectric probe.

61. (ORIGINAL): The method of claim **55** further wherein:

said pin/probe has a point diameter of about the same size as an axon to be moved.

62. (ORIGINAL): The method of claim **55** further wherein:

said pin/probe has a point diameter between about one and ten times a diameter of an axon to be moved.

- 63. (CANCEL):** The method of claim **54** further comprising:
employing a suspending medium for dielectrophoresis and/or electrofusion particularly suited for manipulating axons.
- 64. (CANCEL):** The method of claim **63** further comprising:
adjusting conductivity of a fluidic suspending medium to facilitate DEP.
- 65. (CANCEL):** The method of claim **63** further comprising:
employing a suspending medium with a conductivity of around 200 milli-Siemens/meter (mS/m) or less.
- 66. (CANCEL):** A method of maintaining an axon during manipulation comprising:
employing a CO₂ independent media; and
decreasing the conductivity of the media by diluting a CIM culture media, while maintaining the osmolarity in the physiological range using mannitol and/or a mixture of 8.5% sucrose and 0.3% dextrose.
- 67. (ORIGINAL):** A media for maintaining cell portions while providing desired conductivity for DEP movement comprising:
between 0.5 and 20 % of a standard culture media (e.g., CIM, Gibco/BRL);
a balance being a solution of 5% mannitol in water; and/or
a mixture of 8.5% sucrose and 0.3% dextrose in water.
- 68. (CANCEL):** The method of claim **51** further comprising:
employing a culture media with one or more of the properties of:
able to sustain a physiological PH;
low conductivity;
balance of molarity; and
addition of cell growth factors and/or cell fusion process factors and/or cell repair factors.